

Figure 1:

Amino acid sequences of Cpn60 and Cpn10:

SEQ ID No 1: Cpn10 (encoded by nucleotides pos. 458-751 of Figure 2):

MKIRPLHDRVRRKEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA
VNEGDVVVFGKYSGQNTIDIDGEELLILNESDIYGVLEA

SEQ ID No 2: Cpn60 (encoded by nucleotides pos. 800-2446 of Figure 2):

MAAKDVLFGDSARAKMLVGVNILADAVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
REIELKDKFENMGAQMVKEVASQANDQAGDGTATTATVLAQAIIEGLKSVAAGMNP
MDLKRIGDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKVG
KEGVITVEEGKGLEDELVDVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKKI
DNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVA AVKAPGFGDR
RKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEASV
NTRVDQIRAEIESSTSDYDIEKLQERVAKLGGVAVIKVGAGSEMEMKEKKDRVDDA
LHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQIAGN
AGAEGSVVVDKVKSGTGSFGFNASTGEYGDMIAMGILDPKAVTRSSLQAAASIAGL
MITTEAMVADAPVEEGAGGMPDMGGMGGMGMPGMM

Figure 2:

SEQ ID No 3: DNA coding for Cpn60 and Cpn10:

Cpn10, pos. 458-751

Cpn60, pos. 800-2446

atcaaaaaatgcagcaaggacagattcctgccaagaattagcagaaggtttctttagcactggccggcgcttattattaacgccgg
gtttgtcactgatgcgctgggttttacattactcgtccccgcgacgcgtaaacggttggtccataaggtgattgcatttattaccctc
gcatgatgactgcaagcagctttcaagcgacgggtagtttcaggaaggctcgtttaaatgtacattcgcacactgactcgcaaagca
gtcatgaaaaaatcacaattgaaggcgaatataccaagacgataagtaggtatttttcggctagccgttgaaatcctagtaaaagccc

cgataaattaaccatctatttttcacagaggcaatttagccttgtttaccttattgatcctaatacttgggatccaacagttggagagtctagc
aaatgaaaatccgtccattacatgatcgtattgttgcgcgtaaagaagaagagaccgcaactgcgggtggtatttttacc
ggcgctgcggcagaaaaaccaaataaggtgttattctctgtgggtactggcgtattcttgataatggtcagtgcaagcgctggc
ggftaacgaaggcgatgttgcgttttggtaatactcagggtcaaaatactatcgatcgcgtgaagaattattgattttgaatga
aagtatactacggcggttttagaagcttaattattacactcacttttttatttaacctacaaaatttaaggaaagatcatggctgctaaagacg
tattatttggatagcgcacgcgcaaaaatgttggtaggtgtaaacattttagccgacgcagtaagagttaccttaggacctaa
aggctgtaacgttgttatagaaaaatcatttgggtgcaccgatcatcaccaaagatggtgtttctgttgcgcgtgaaatcgaattgaaagaca
aattcgaataacatggcgccacagatggttaagggaaggtgttctcaagccaacgaccaagccggtgacggcacaacgacagcgactg
tactagcacaggcgattatcagcgaaggcttgaatatctgttgcggctggcatgaatccaatggatcttaaacgttggtattgataaagctac
ggctgctgttgttgcgcgcaataaagaacaagctcagccttgccttgatacaaaagcaatcgctcaggtagggacaatctctgccaatgc
cgatgaaacgggttgctgtttaaattgctgaagcgatggaaaaagtcggtaaaagaaggtgtgattaccgttgaagaaggcaaaaggccttg
aagacgagcttgatgtttagaaggcatgcagttcgatcgcggttacttgtctccgtacttcatcaacaaccaagaaaaaatgaccgtag
aaatggaaaatccattaattctatttgggtgataaaaaattgataacctcaagagctgttgcgaattcttgaaaacgtcgtctaaatcaggtc
gtccattattgatcgttgcgtgaagatgttgaaggccaagcactagcaacattggtagtaaacacttgcgcggcacattcaaggttgc
agcggtaaaagcccctgggttggcgatcgtcgtaaagcgatgttgaagatcttgccatcttgacgggtggtcagggtatttctgaagag
ctagggtgctttagaaaactgcggatccttcttcttgggtacggcaagcaaggtgttatcgataaagaaaacaccgtgattgttga
tggcgagggtactgaagcaagcgtaatactcgtgttgaccagatccgtgctgaaatcgaaagctcgacttctgattacgacatcgaaaa
gttacaagaacgcgttgctaagcttgcggcgggcgttgcgtgattaaaggttgggtgcgggttctgaaatggaaatgaaagagaagaaa
gaccgtgttgacgatgcacttcatgcaactcgcgcagcgggtgaagaaggtgttgttgcgggtggtgtgttgccttgatcgcgcactct
cttcagtaaccgttgttgggtgataacgaagatcaaaacgtcgggtattgcattggcaccttctgctgatggaagctcctatccgtcaaatcgc
gggtaacgcagggtgctgaagggtcagtggtgttgataaaagtgaatctggcacaggtagcttgggttttaacccagcagcaggtgagt
atggcgatatgattgcgatgggtatttttagacctgcaaaagtcacgcgttcatctctacaagccgcggcgtctatcgagggttggatgat
cacaaccgaagccatggttgcggatgcgcctgttgaagaagcgctgggtggtatgcctgatatggcgggcatgggtggaatggcggg
tatgcctggcatgatgtaatcacttgtgattcattgtcctgatctgcttaccgtgtaaaaagatcagggtcaaggctgtctctataaaaagcc
gtatcttggatgagtggtgtcttctgtgtaaaacgacattcttgagtgccgcttttttatttgggtcataaaattcagaatattgtgtaatttta
tgtaactagctggcctataatgttgagttcctctgggtggcatgatctcatggtacttcaactaagcctgattcactgcg
gctttaacagtaaaataataacgcaacgtagaacaataaagcgtatggcattaatgaagacggctgcatttaattcagatc

Figure 3:

SEQ ID No 4: Amino acid sequence of esterase cloned from *Oleispira antarctica* (EstRB8):

EstRB8 (encoded by nucleotides 1145 to 2143 Frame 2 of Figure 4) 333 aa

MKNTLKSSSRFSLKQLGTGALISSLFFGGCTTTQQDNLYTGVM SLARDSAGLEVKTA
SAGDVNLTYMERQGSDDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG
HGDSEQLLTDDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGG AISAIYSLSHPEKVKSL
TLIDAAGVDGDTESEYYKVLAEKGK NPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL
RKTLARAEINN KIFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD
VSAAA AFKKIIPQATVHIFPEVGHLPMVEIPSES AKVYEEFLSSIK

Figure 4:

SEQ ID No 5: DNA fragment from plasmid pBK1Est coding for esterase of *Oleispira antarctica* (EstRB8):

Nucleotide positions 1-100 correspond to reverse complement of positions 1196-1121 and 3799-3939 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene).

Positions 101-105 are *Bam*HI – *Sau*3A1 fusion and positions 3795-3798 are *Sau*3A1-*Bam*HI-fusion.

acaggaaacagctatgacctgattacgccaagctcgaaattaaccctcactaaagggaacaaaagctggagctcgcgccctgcag
gtcgacactagtggatcaacggcgttcattggtactggctgagctcagcgtcataatgccgatgcgatactggcgcgtactgagctact
tctctgctagcaccgattttctaatagcgcagcttcttttattctgaacgggcaactgatgtagtttttactaaccggccttttaggcatgg
taaactcttcgatattcaaaattattactgttcataattacaatcatagctacaggctagaggcccaaaattgcagctgatattcacctttatttc
taagcattattacactcatcgcggtgttattaattgtgctaaataaaaatacccgtagcggaaaaattcagcaaatagccaaagaaaacga
ttggcaataccaagaattcatcgattttgatgatgacattaagcaggcaaaactttggcctattaaactacagtcaaaatgcaatttttagacat
ctcattcaagcaactgacgaacactatggcttagcgtttaagacctttgactgtcgagcgttagaaccttcaggtattcacaatagcagcttt
atthttattaccctcgactaaagactgaattcaataacctacacatttgccttaagtcgacatattcaagataaagatgccttcactgacatca
gtcaccaacaatcaatcaaacaccaataccaatcgcaaaaactcataaaactagccgatcaccaatcccaaaagcgttcaaaaatgaa
acgagcacgtcacacaaaatcaatttatacgtaacgaaccagggtcaaaacttatcggtttttgagcacgtttgtccactaatgaaagaga
aaagtcgttaattcactggccttttggcgtatccgcaccttcacatagaaattagtaattggcatgctactggcctttaaaaagaatcagtttaatt
gaagaaacctcgcttatctcagccattaccgctgtagccgaatttgcgcttatcctcagccatgattaaactgacgccaatataataagac
atactaattaataactcccttaattgagaagaataatgaaaacacactcaaatcctcatcacgttttagctgaaacaactcggcaccggc
gctctgattatctccagtttgttctcgggtgtgcaccacaacacaagaataattatacacaggggttatgtctcttgcgagagacagc
gctggcctagaagftaaaacagcctctgccggtgacgtcaatcttacttatatggaacgccaaaggcagtgacaaagataatgccgaaag

cgttattttattacacggtttctctgctgataaagataactggattctttttaccaaaagaattcgaataaaatcatgttatcgctgctgattta
gcgggacatggcgattcagaacaattattaacgactgattacgggtctataaaacaagccgagcgttagatatcttcttctggttagg
ggttaactcatttcacatcgccggttaattcaatggggggggctatcagcgaatctacagtttagtcaccagagaaagttaaaagtctt
acattgatcgatgcagcaggtgctgatggcgatactgaaagcgaatactacaaagtttggcagaaggaagaatcctttaattgcaact
gatgaagcaagtttgaataccgcatgggtttccatgactcagcctctttctaccttggccactaagaccttctttattacgtaaaacg
ctagcccgtgccgagatcaataacaaaatttttccgatgctgaaaaccaaagaacgtttaggaatgactaacttcaacagaaaattg
aagtgaatggctcaacatccattgccaacactgattatgtggggcaaagaagatcgcttcttgacgtatccgcagcagcggccttc
aaaaaataattccacaagcaactgttcataatcttctgaagtaggccacctacatggttagaaattcctagtgaagcgctaaagtatt
gaagagttttgtctctattaaataagagcacataatcatgactgacttataaacagccaagcatttaaatgcttggctgtttatattaatgg
ccaaattattcaacgaccaagctctgcggtaaaatcgagtggttcttgttttcatcaacagcaacaaacgtgaataccccgtaatcg
cattttctgattatcaaaatacatactttccaccagcatattaacttcaacttttaactcgtccgccctacctctataacactggcagtcatt
cgacaatggtacctgcgggaacaggatgcttaaaatcgattcgatcactgctgacggttacgatgctttgtcgagaaaaacgagtcgct
gcaataaaagaaacctcatccactgcattgcagtgccaccgaataacgtaicgatgatgttgtgtctctggaaataccgctttaga
aatagtggttttgatacgegtttcgtgcgaataatatcttctctgctaagagttgcggatggcatacataaaactcgttgattaagatta
ataataaatagttaacagtataattgaactgagggctgaagaactctaatacctctgaagaactttgagggcgctagagagaaaagacca
gtgataatattcatcttgccatgagagcttatcatgaaagcctgtgcttaaaatcaatcattatatttattcatctttaattgaaataataccaat
atatttcatatataatttcacactaccttatecactagacttcccgcgataggcgcaacaatacaacgcaagttcacaataaagcgggttc
gctgcaacacatgccctagcgtctaaagtagcacgcacaacactggccagtcgtactagcccccttgcgattcgtgcagacgagcaac
aagcgtatttaaaacttacctaaatttctaaccaccaccattggttctttccacaaactcaaaaaactcgtcaaatccgcttgcaatttaaacg
cgatgacatagatctaatecgattatcaaacccgattcaagcgtcattaaaaacgcaccactggcaagaagttctacctgcactgacca
atatgcaagcggcgggcggaagagctgcctttgatcatcaagaagaaggagcagcaaaaggagaaaacaatcaaaaagaggagag
caatcaataaaaaacgagttattgaggattttaattttaaaacaggtatattaataccctctctctagtaaacatgactgtatttacaaaa
ataaatagaggtataccatgtcaaacatctggtttgaagtaccaaagattgaagtattaaaccgtcaaatggaaaaactgcctgcagcaa
cttaggcattcaaaattacagaaattggcgatgattatcactggcacaatgccagcagatgcacgtaccttcagccaatgggactgatt
catggcggtcacaatgtattgtctggcagaaacactgggcagcatggcagctaactgctgtatttaattgtctcaagaatattgtgttgcc
aagaaatfaacgccaaccacatagcgggtgttctccggcatagtgactggcacagcaacgctagtacacaaaggaagaacctccca
gatttgggaaattcgcatcgttaacgatccaaagaattcaaaaagcttctcgagagtacttctagagcgggccgccccatcgattttcc
accgggtgggggtaccaggttaagtgtacccaattcgcctatagtgagtcgtattacaattcactggccgtcgttttac

Figure 5:

Amino acid sequences expressed from vector pBK1CpnEst: - the co-expression of fragments encoding native chaperonines with the esterase gene (EstRB8), all from *Oleispira antarctica*

SEQ ID No 6: cpn10 (nucleotides 113 to 403: Frame 2 of Figure 6) 97 aa:

MKIRPLHDRVRRKEEETATAGGILPGAAAEKPNQGVVISVGTGRILDNGSVQALA
VNEG DVVVF GKYS GQNTIDIDGEELLILNESDIYGVLEA

SEQ ID No 7: cpn60 (nucleotides 455 to 2098: Frame 2 of Figure 6) 548 aa:

MAAKDVLFGDSARAKMLVGVN ILADAVRVT LGPKGRNVVIEKSFGAPIITKDGVSV A
REIELKDKFENMGAQM VKEVASQANDQAGDGT TATVLAQAIISEGLKSVAAGMNP
MDLKR GIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANA DETVGR LIAEAMEKVG
KEGVITVEEGKGLEDEL DVVEGMQFDRGYLSPYFINNQE KMTVEMENPLILLVDK KI
DNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVA AVKAPGFGDR
RKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEASV
NTRVDQIRAEIESSTSDYDIEKLQERVA KLAGGVAVIKVGAGSEMEMKEKKDRVDDA
LHATRAAVEEGVVAGGGVALIRALSSVTTVVGDNEDQNVGIALALRAMEAPIRQIAGN
AGAEGSVVVDKVKSGTGSFGFNASTGEYGD MIAMGILDPK VTRSSLQAAASIAGL
MITTEAMVADAPVEEGAGGMPDMGGMGGMGGMGMPGMM

SEQ ID No 8: estRB8 (nucleotides 2579 to 3577: Frame 2 of Figure 6) 333 aa:

MKNTLKSSSRFSLKQLGTGALISSLFFGGCTTTQQDNLYTGVM SLARDSAGLEVKTA
SAGDVNLTYMERQGS DKDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG
HGDSEQLLT TDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGG AISAIYSLSHPEKVKSL
TLIDAAGVDGDTESEYYKVLAEGKNPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL
RKTLARAEINN KIFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD
VSAAA AFKKIIPQATVHIFPEVGHLP MVEIPSESAKVYEEFLSSIK

Figure 6:

SEQ ID No 9: pBK1CpnEst: - the fusion of native chaperonine-coding fragments with
esterase of *Oleispira antarctica* (EstRB8)

The DNA fragment coding for Cpn10 and Cpn60 is flanked by *Sac*I site (pos. 69-75) and *Sal*I site (encoded by pos. 2138-2143 of Figure 7):

Nucleotide positions 1-75 correspond to reverse complement of positions 1196-1121 and positions 5233-5273 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene)

Small letters – the Cpn10-Cpn60 encoding fragment,

Capital italics – fragments of vector pBK-CMV

Capital letters – fragment coding for EstRB8 from plasmid pBK1Est

ACAGGAAACAGCTATGACCTTGATTACGCCAAGCTCGAAATTAACCCTCACTAAAGGGA
*ACAAAAGCTGGAGCTC*ctaataactgggatccaacaggttgagagcttagcaaatgaaaatccgtccattacatgatcgtatt
gttcttcgccgtaaagaagaagagaccgcaactgcgggtgggtattttaccgggcgtgcggcagaaaaacaaatcaagggtgtgt
tatctctgtgggtactggccgtattcttgataatgggtcagtgaacgcgtggcggttaacgaaggcgatgtgtcgttttgtaaatactc
agggtcaaaatactatcgatcgcgttggaagaattatgtatttgaaatgatatctacggcggtttagaagcttaattattacactca
ctttttatttaacctacaaaatttaaggaaagatcatggcgtctaaagacgtattatttgggtgatagcgacgcgcaaaaatgttggttaggt
gtaaacatttttagccgacgcagtaagagttaccttaggacctaaaggctgtaacgttggttatagaaaaatcatttgggtgcaccgatcatcac
caaagatggtgtttctgttcgcgtgaaatcgaattgaaagacaaatcgaaaacatggcgccagatggttaaggaggtgcttctca
agccaacgaccaagccggtgacggcacaacgacagcgactgtactagcacaggcgattatcagcgaaggctgaaatctgttgccg
ctggcatgaatccaatggatcttaaacgttggtattgataaagctacggctgctgtgttgccgccattaaagaacaagctcagccttgcttg
gatacaaaagcaatcgctcaggtagggacaatctctgccaatgccgatgaacgggttggtcgttaattgctgaagcgatggaaaaagt
cggtaaagaagggtgtgattaccgttgaagaaggcaaaggccttgaagacgagcttgatgttgtagaaggcatgcagttcgatcgcggtt
actgtctccgtacttcatcaacaaccaagaaaaatgaccgtagaaatgaaaaatcatttaattctattgggtgataagaaaattgataac
cttcaagagctgttgccaattcttgaaaacgtcgctaaatcaggtcgctccattattgatcgttgctgaagatgttgaaggccaagcactagc
aacattggtagtaacaacttgcgcggcacattcaagggtgcagcggttaaaagcccctgggtttggcgatcgctgtaaagcgatgttgca
agatcttgccatcttgacgggtggtcagggtatttctgaagagctagggtatgcttttagaaactgcggatccttcttcttgggtacggcaag
caagggtgttatcgataaagaaaacaccgtgattgttgatggcgagggtactgaagcaagcggttaatactcgtgttgaccagatccgtgct
gaaatcgaaagctcgactctgattacgacatcgaaaagttacaagaacgcgttgctaagcttgcggcgccgttgccgtgattaagggt

gggtgcgggttctgaaatggaatgaaagagaagaagaccgtgttgacgatgcacttcacgaactcgcgcagcgggtgaagaagg
gttgttcgggtgggtgttgccttgattcgcgcactctcttcagtaaccgtgttggtgataacgaagatcaaacgcgggtattgcattg
gcacttcgtgcgatggaagctcctatccgtcaaatcgcgggtaacgcaggtgctgaagggtcagtggtgttgataaagtgaatctgg
cacaggtagctttgggttaacgccagcacaggtgagtatggcgatatgattgcgatgggtattttagaccctgcaaaagtcacgcgttca
tctctacaagccgcggcgtctatcgcaggtttgatgatcacaaccgaagccatgggtgcggatgcgcctgttgagaaggcgtgggtg
tatgcctgatatgggcccgtgggtggaatgggcgggtatgcctggcatgatgtaatacactttgtgattcattgtcctgatctgcttaccgtG
TCGACATATTCAAGATAAAGATGCCTTCACTGACATCAGTCACCAACAATCAATC
AAACACCAATACCAATCGCAAAAACCTCATAAACTAGCCGATCACCAAAATCCCA
AAAGCGTTCAAAAATGAAACGAGCACGTCACACAAAATCAATTTATACGCTAAC
GAACCAGGTCAAACCTTATCGTTTTTTTGAGCACGTTTGTTCCTACTAATGAAAGAG
AAAAGTCGTTAATTCCTGACTGGCTTTTGGCGTATCCGCACCTTCACATAGAAATTAGT
AATGGCATGCTACTGGCCTTTAAAAAGAATCAGTTAATTGAAGAAACCTCGCTTA
TCTCAGCCATTACCGCTGTAGCCGAATTTGCGCTTATCCTCAGCCATGATTAACT
GACGCCAATTAATATAAGACATACTAATTAATAACTCCCTTAATTGAGAAGAATA
ATGAAAAACACACTCAAATCCTCATCACGTTTTAGTCTGAAACAACCTCGGCACCG
GCGCTCTGATTATCTCCAGTTTGTCTTTCGGTGGTTGCACCACAACACAAGAT
AATTTATACACAGGGGTATGTCTCTTGCGAGAGACAGCGCTGGCCTAGAAGTTA
AAACAGCCTCTGCCGGTGACGTCAATCTTACTTATATGGAACGCCAAGGCAGTGA
CAAAGATAATGCCGAAAGCGTTATTTTATTACACGGTTTCTCTGCTGATAAAGAT
AACTGGATTCTTTTTACCAAAGAATTCGATGAAAAATATCATGTTATCGCTGTCGA
TTTAGCGGGACATGGCGATTGAGAACAATTATTAACGACTGATTACGGTCTCATA
AAACAAGCCGAGCGTTTAGATATCTTCTTATCTGGCTTAGGGGTAACTCATTTC
CATCGCCGGTAATTCAATGGGGGGGGCTATCAGCGCAATCTACAGTTTGAGTCAC
CCAGAGAAAGTTAAAAGTCTTACATTGATCGATGCAGCAGGTGTCGATGGCGATA
CTGAAAGCGAATACTACAAAGTTTTGGCAGAAGGTAAGAATCCTTTAATTGCAAC
TGATGAAGCAAGTTTTGAATACCGCATGGGTTCACCATGACTCAGCCTCCTTTCC
TACCTTGGCCACTAAGACCTTCTTTATTACGTAACGCTAGCCCGTGCCGAGATC
AATAACAAAATTTTTTCCGATATGCTGAAAACCAAAGAACGTTTAGGAATGACTA
ACTTTCAACAGAAAATTGAAGTGAAAATGGCTCAACATCCATTGCCAACACTGAT
TATGTGGGGCAAAGAAGATCGCGTTCTTGACGTATCCGCAGCAGCGGCCTTCAAA
AAAATAATTCCACAAGCAACTGTTTCATATTTTTCTGAAGTAGGCCACCTACCTAT
GGTAGAAATTCCTAGTGAAAGCGCTAAAGTTTATGAAGAGTTTTTGTCTCTATTA
AATAAGAGCACATAATCATGACTGACTTATAAACAGCCAAGCATTAAAAATGCTT
GGCTGTTTATTTAATGGCCAAATTATTCAACGACCAAGCTCTGCGGTAAAATCG

CAGTGGGTTTCTTGTTTTTCATCAACAGCAACAAACGTGAAATACCCCGTAATCGC
ATTTTTCTGATTATCAAAATACATACTTTCCACCAGCATATTAACCTTCAACTTTTA
AACTCGTCCGCCCTACCTCTATAACACTGGCAGTCAATTTCGACAATGGTACCTGC
GGGAACAGGATGCTTAAAATCGATTTCGATCACTGCTGACGGTTACGATGCTTTGT
CGAGAAAAACGAGTCGCTGCAATAAAAGAAACCTCATCCATCCACTGCATTGCA
GTGCCACCGAATAACGTATCATGATGATTTGTTGTCTCTGGAAATACCGCTTTAGA
AATAGTGGTTTTTGATACGCGCTTTCGCTGCGCAATAATATCTTCTCTGCTAAGAG
TTGCGGATGGCATACTAACTCGCTTGATTAAGATTAATAATAAATAGTTAACA
GTATATTGAACTGAGGGTCTGAAGAACTCTAATACCTCTGAAGAACTTTGAGGCC
GCTAGAGAGAAAAGACCAGTGATAATATTTTCATCTTGCCATGAGAGCTTATCATG
AAAGCCTGTGCTTAAAATCAATCATTATATTTATTCATCTTTAATTGAAATAATAC
CAATATATTTTCATATATAATTTACACTACCCTTATCTCACTAGACTTCCCGCGCA
TAGGCGCAAACAATCAACGCAAGTTCACAATAAAGCGGTTTCGCTGCAACACATG
CCCTAGCGTCTAAAGTAGCACGCACAACACTGGCCAGTCGTAAGTACCTTTTGC
GATTCGTGCAGACGAGCAACAAGCGCTATTAACTTACCTAAATTTCTAACCACC
ACCATTGGTTCTTTTCCACAACTCAAAAACTCGTCAAATCCGCTTGCAATTTAA
ACGCGATGACATAGATCTAATCGATTATCAAACCCGCATTCAAGCGCTCATTAAA
AACGCACCACTGGCAAGAAGTTCTACCTGCACTGACCAATATGCAAGCGGCGGC
GGAAGAGCTGCCTTTGATCGATCAAGAAGAAGGGAGCAGCAAAGAGGAAAACA
ATCAAAAAGAGGAGAGCAATCAAATAAAAACGAGTTATTGAGGATTTTAATTTTA
AAACAGGTATATTAATACCCTCTCTCGTAGTAAACAATGACTGTATTTACACAAA
AATAAATAGAGGTATACCATGTCAAACATCTGGTTTGAAGTACCAAAGATTGAAG
TATTAAACCGTCAAATGGAAAATACTGCCTGCAGCAACTTAGGCATTCAAATTAC
AGAAATTGGCGATGATTATATCACTGGCACAATGCCAGCAGATGCACGTACCTTC
CAGCCAATGGGACTGATTCATGGCGGCTCAAATGTATTGCTGGCAGAAACACTGG
GCAGCATGGCAGCTAACTGCTGTATTAATTTGTCTCAAGAATATTGTGTTGGCCA
AGAAATTAACGCCAACCACATACGCGGTGTTTCGTTCCGGCATAGTGACTGGCACA
GCAACGCTAGTACACAAAGGAAGAACCTCCCAGATTTGGGAAATTCGCATCGTTA
ACGATCCAAAGAATTCAAAAAGCTTCTCGAGAGTACTTCTAGAGCGGCCGCGGGCCCA
TCGATTTTCCACCCGGGTGGGGTACCAGGTAAGTGTACCCAATTCGCCCTATAGTGAGT
CGTATTACAATTCCTGGCCGTCGTTTTAC

Figure 7:

Amino acid sequences expressed from vector pBK1CpnSREst: - the co-expression of the stabilized single ring mutant chaperonin with the esterase gene (EstRB8) from *Oleispira antarctica* (cpn10::stabilized single ring mutant

Glu461Ala/Ser463Ala/Val464AlaGlu460Ala/Ser462Ala/Val463Ala::est)

SEQ ID No 10: cpn10 (nucleotides 113 to 403: Frame 2 of Figure 8) 97 aa:

MKIRPLHDRVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA
VNEGDVVVFVKYSGQNTIDIDGEELLILNESDIYGVLEA

Below – ***Capital bold letters*** are the mutations introduced

SEQ ID No 11: stabilized single ring mutant of cpn60 (nucleotides 455 to 2098: Frame 2 of Figure 8) 548 aa:

MAAKDVLFGDSARAKMLVGVNILDVVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
REIELKDKFENMGAQMVKEVASQANDQAGDGTATVLAQAIISEGLKSVAAGMNP
MDLKRIGDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKVG
KEGVITVEEGKGLEDELVDVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKKI
DNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVAAVKAPGFGDR
RKAMLQDLAILTGGQVISEELGMSLETADPSSLGTSKVVVIDKENTVIVDGAGTEASV
NTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVDDA
LHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQIAGN
AGA**AGAA**VVDKVKSGTGSFGFNASTGEYGDMIAMGILDPKAVTRSSLQAAASIAGL
MITTEAMVADAPVEEGAGGMPDMGGMGGMGGMPGMM

SEQ ID No 12: EstRB8 (nucleotides 2579 to 3577: Frame 2 of Figure 8) 333 aa:

MKNTLKSSSRFSLKQLGTGALHSSLFFGGCTTTQQDNLYTGVM SLARDSAGLEVKTA
SAGDVNLTYMERQGSDDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG
HGDSEQLLTDDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGGAIASISLHPEKVKSL

TLIDAAGVDGDTSEYYKVLAEKGKPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL
RKTLARAEINNKFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD
VSAAAFAFKIIPQATVHIFPEVGHLPMEIPSESAKVYEEFLSSIK

Figure 8:

SEQ ID No 13: DNA sequence of vector pBK1CpnSREst: the expression cassette for the co-expression of the stabilized single ring mutant chaperonin with the esterase gene (EstRB8) from *Oleispira antarctica* (cpn10::stabilized single ring mutant Glu461Ala/Ser463Ala/Val464AlaGlu460Ala/Ser462Ala/Val463Ala::est)

Nucleotide positions 1-75 correspond to reverse complement of positions 1196-1121 and positions 5233-5273 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene)

DNA fragment coding for Cpn10 and Cpn60 is flanked by *SacI* site (pos. 69-75) and *SalI* site (pos. 2138-2143).

In the DNA sequence:

Small letters – the Cpn10-Cpn60 coding fragment,

Capital italics – fragments of vector

Capital letters – fragment coding for EstRB8 from plasmid pBK1Est

Capital bold letters = introduced mutations

ACAGGAAACAGCTATGACCTTGATTACGCCAAGCTCGAAATTAACCCTCACTAAAGGGA
ACAAAAGCTGGAGCTCtaataacttgggatccaacagttggagagcttagcaaatgaaaatccgtccattacatgatcgtatt
gtgttcgccgtaaagaagaagagaccgcaactgcgggtggtattttaccggcgctgcggcagaaaaacaaatcaaggtgtgt
tatctctgtgggtactggccgtattcttgataatggttcagtgcaagcgctggcggttaacgaaggcgatgtgtcgttttggtaaatactc
aggtaaaaatactatcgatcgtatggaagaattatgtatttgaatgaaagtatatctacggcggtttagaagcttaattattacactca
ctttttatttaacctacaaaatttaaggaaagatcatggctgctaaagacgtatttttggtgatagcgacgcgcaaaaatgttgtaggt
gtaaacatttttagccgacgcagtaagagttaccttaggacctaaaggcgtgaacgtgttatagaaaaatcatttggtgcaccgatcatcac
caaagatggtgtttctgttgcgcgtgaaatgaattgaaagacaaatcgaaaacatgggcgcacagatggttaagggaagtgcttctca
agccaacgaccaagccggtgacggcacaacgacagcgactgtactagcacaggcgattatcagcgaaggcttgaatctgttgcgg

ctggcatgaatccaatggatcttaaacgtggattgataaagctacggctgctgtgttgccgccattaaagaacaagctcagccttgcttg
gatacaaaagcaatcgctcaggtagggacaatctctgccaatgccgatgaaacgggtggctgttaattgctgaagcgatggaaaaagt
cggtaaagaaggtgtgattaccgtgaagaaggcaaaggcctgaagacgagcttgatgtttagaaggcatgcagttcgatcgcggtt
actgtctccgtacttcatcaacaaccaagaaaaatgaccgtagaatggaaaatccattaattctattggttgataagaaaattgataac
cttcaagagctgttgccaattcttgaaaacgtcgctaaatcaggtcgccattattgatcggtgctgaagatgttgaaggccaagcactagc
aacattggtagtaaacaacttgcgcggcacattcaaggttgacgcgggttaaagccccctgggtttggcgatcgctgtaaagcgatgttgca
agatcttgccatcttgacgggtggtcaggttatttctgaagagctagggatgtctttaaaaactgcggatccttcttcttgggtacggcaag
caaggttggtatcgataaagaaaacaccgtgattgttgatggcgaggtactgaagcaagcgtaataactcggttgaccagatccgtgct
gaaatcgaaagctcgacttctgattacgacatcgaaaagttacaagaacgcgttgctaagcttgcggcgcggttgccgtgattaaggtt
gggtgcgggttctgaaatggaaatgaaagagaagaagaccgtgttgacgatgcacttcatgcaactcgcgcagcgggtgaagaaggt
gttgttcgggtgggtgtgtgttcttgcgcactctcttcagtaaccgttgttgggtgataacgaagatcaaaacgtcggtattgcattg
gcacttctgcatggaagctcctatccgtcaaatcgcgggtaacgcaggtgctgCagggGcagCggtgttgataaagtgaatct
ggcacaggtagcttgggtttaacgccagcacaggtgaglatggcgatgatgcatgggtatttagacctgcaaaagtcacgcgtt
catctctacaagccgcggcgctctatcgaggtttagatgacacaaccgaagccatggttgcggatgcgcctgttgaagaaggcgctggt
ggatgcctgatatggcgggcatgggtggaatggcggtatgccggcatgatgtaatcacttgtgattcattgtctgatctgctaccg
tGTGACATATTCAAGATAAAGATGCCTTCACTGACATCAGTCACCAACAATCAAT
CAAACACCAATACCAATCGCAAAAACCTCATAAACTAGCCGATCACCAAATCCC
AAAAGCGTTCAAAAATGAAACGAGCACGTACACAAAATCAATTTATACGCTAA
CGAACCAGGTCAAACCTTATCGTTTTTTTGAGCACGTTTGTTCCTACTAATGAAAGAG
AAAAGTCGTTAATTCCTGACTGGCTTTTGGCGTATCCGCACCTTCACATAGAAATTAGT
AATGGCATGCTACTGGCCTTTAAAAAGAATCAGTTAATTGAAGAAACCTCGCTTA
TCTCAGCCATTACCGCTGTAGCCGAATTTGCGCTTATCCTCAGCCATGATTAACT
GACGCCAATTAATATAAGACATACTAATTAATAACTCCCTTAATTGAGAAGAATA
ATGAAAAACACACTCAAATCCTCATCACGTTTTAGTCTGAAACAACCTCGGCACCG
GCGCTCTGATTATCTCCAGTTTGTCTTCGGTGGTTGCACCACAACACAACAAGAT
AATTTATACACAGGGGTATGTCTCTTGCGAGAGACAGCGCTGGCCTAGAAGTTA
AAACAGCCTCTGCCGGTGACGTCAATCTTACTTATATGGAACGCCAAGGCAGTGA
CAAAGATAATGCCGAAAGCGTTATTTTATTACACGGTTTCTCTGCTGATAAAGAT
AACTGGATTCTTTTTACCAAAGAATTCGATGAAAAATATCATGTTATCGCTGTCGA
TTTAGCGGGACATGGCGATTGAGAACAATTATTAACGACTGATTACGGTCTCATA
AAACAAGCCGAGCGTTTAGATATCTTCTTATCTGGCTTAGGGGTAACTCATTCA
CATCGCCGTAATTCAATGGGGGGGGCTATCAGCGCAATCTACAGTTTGAGTCAC
CCAGAGAAAGTTAAAAGTCTTACATTGATCGATGCAGCAGGTGTCGATGGCGATA
CTGAAAGCGAATACTACAAAGTTTTTGGCAGAAGGTAAGAATCCTTTAATTGCAAC

TGATGAAGCAAGTTTTGAATACCGCATGGGTTTCACCATGACTCAGCCTCCTTTCC
TACCTTGGCCACTAAGACCTTCTTTATTACGTAAAACGCTAGCCCGTGCCGAGATC
AATAACAAAATTTTTCCGATATGCTGAAAACCAAAGAACGTTTAGGAATGACTA
ACTTTCAACAGAAAATTGAAGTGAAAATGGCTCAACATCCATTGCCAACACTGAT
TATGTGGGGCAAAGAAGATCGCGTTCTTGACGTATCCGCAGCAGCGGCCTTCAAA
AAAATAATTCCACAAGCAACTGTTTCATATTTTTCTGAAGTAGGCCACCTACCTAT
GGTAGAAATTCCTAGTGAAAGCGCTAAAGTTTATGAAGAGTTTTTGTCTCTATTA
AATAAGAGCACATAATCATGACTGACTTATAAACAGCCAAGCATTTAAAATGCTT
GGCTGTTTATTTTAATGGCCAAATTATTCAACGACCAAGCTCTGCGGTAAAATCG
CAGTGGGTTTCTTGTTTTTCATCAACAGCAACAAACGTGAAATACCCCGTAATCGC
ATTTTTCTGATTATCAAAATACATACTTTCCACCAGCATATTAACCTTCAACTTTTA
AACTCGTCCGCCCTACCTCTATAACACTGGCAGTCAATTCGACAATGGTACCTGC
GGGAACAGGATGCTTAAAATCGATTTCGATCACTGCTGACGGTTACGATGCTTTGT
CGAGAAAAACGAGTCGCTGCAATAAAAGAAACCTCATCCATCCACTGCATTGCA
GTGCCACCGAATAACGTATCATGATGATTGTTGTCTCTGGAAATACCGCTTTAGA
AATAGTGGTTTTTGATACGCGCTTTCGCTGCGCAATAATATCTTCTCTGCTAAGAG
TTGCGGATGGCATAACATAAACTCGCTTGATTAAAGATTAATAATAAATAGTTAACA
GTATATTGAACTGAGGGTCTGAAGAACTCTAATACCTCTGAAGAACTTTGAGGCC
GCTAGAGAGAAAAGACCAGTGATAATATTTTCATCTTGCCATGAGAGCTTATCATG
AAAGCCTGTGCTTAAAATCAATCATTATATTTATTCATCTTTAATTGAAATAATAC
CAATATATTTTCATATATAATTTACACTACCCTTATCTCACTAGACTTCCCGCGCA
TAGGCGCAAACAATCAACGCAAGTTCACAATAAAGCGGTTCGCTGCAACACATG
CCCTAGCGTCTAAAGTAGCACGCACAACACTGGCCAGTCGTACTAGCCCCTTTGC
GATTCGTGCAGACGAGCAACAAGCGCTATTAAACTTACCTAAATTTCTAACCACC
ACCATTGGTTCTTTTCCACAACTCAAAAACTCGTCAAATCCGCTTGCAATTTAA
ACGCGATGACATAGATCTAATCGATTATCAAACCCGCATTCAAGCGCTCATTAAA
AACGCACCACTGGCAAGAAGTTCTACCTGCACTGACCAATATGCAAGCGGCGGC
GGAAGAGCTGCCTTTGATCGATCAAGAAGAAGGGAGCAGCAAAGAGGAAAACA
ATCAAAAAGAGGAGAGCAATCAAATAAAAACGAGTTATTGAGGATTTTAATTTTA
AAACAGGTATATTAATACCCTCTCTCGTAGTAAACAATGACTGTATTTACACAAA
AATAAATAGAGGTATACCATGTCAAACATCTGGTTTGAAGTACCAAAGATTGAAG
TATTAAACCGTCAAATGGAAAATACTGCCTGCAGCAACTTAGGCATTCAAATTAC
AGAAATTGGCGATGATTATATCACTGGCACAATGCCAGCAGATGCACGTACCTTC
CAGCCAATGGGACTGATTCATGGCGGCTCAAATGTATTGCTGGCAGAAACACTGG

GCAGCATGGCAGCTAACTGCTGTATTAATTTGTCTCAAGAATATTGTGTTGGCCA
AGAAATTAACGCCAACCACATACGCGGTGTTTCGTTCCGGCATAGTGACTGGCACA
GCAACGCTAGTACACAAAGGAAGAACCTCCCAGATTTGGGAAATTCGCATCGTTA
ACGATCCAAAGAATTCAAAAAGCTTCTCGAGAGTACTTCTAGAGCGGCCGCGGGCCCA
TCGATTTTCCACCCGGGTGGGGTACCAGGTAAGTGTACCCAATTCGCCCTATAGTGAGT
CGTATTACAATTCAGTGGCCGTCGTTTTAC

Figure 9:

Amino acid sequence of the stabilized single ring mutant

Glu461Ala/Ser463Ala/Val464AlaGlu460Ala/Ser462Ala/Val463Ala of Cpn60:

SEQ ID No 14: Cpn10 (nucleotides 458-751 of Figure 10):

MKIRPLHDRVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA
VNEG DVVVF GKYS GQNTIDIDGEELLILNESDIYGVLEA

SEQ ID No 15: Cpn60 (nucleotides 458-751 of Figure 10):

MAAKDVLFGDSARAKMLVGVN ILADAVRVT LGPKGRNVVIEKSFGAPIITKDGVSV
REIELKDKFENMGAQMVK EVASQANDQAGDGT TTVLAQAIISEGLKSVAAGMNP
MDLKR GIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANA DETVGR LIAEAMEKVG
KEGVITVEEGKGLEDEL DVVEGMQFDRGYLSPYFINNQE KMTVEMENPLILLVDK KI
DNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVA AVKAPGFGDR
RKAMLQDLAILTGGQVISEELGMSLETADPSSLG TASKVVIDKENTVIVDGAGTEASV
NTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVDDA
LHATRAAVEEGVVAGGGVALIRALSSVTTVVGDNEDQNVGIALALRAMEAPIRQIAGN
AGAAGAAVVDKVKSGTGSFGFNASTGEYGD MIAMGILDPAKVTRSSLQAAASIAGL
MITTEAMVADAPVEEGAGGMPDMGGMGGMGGMGMPGMM

Figure 10:

SEQ ID No 16: DNA sequence of the stabilized single ring mutant

Glu461Ala/Ser463Ala/Val464AlaGlu460Ala/Ser462Ala/Val463Ala:

In the DNA sequence:

Small letters – the Cpn10-Cpn60 coding fragment,

Big bold letters = introduced mutations

atcaaaaaatgcagcaaggacagattcctgcccagaattagcagaagggttcttgtagcactggccggcgctttattattaacgccgg
gtttgtcactgatgcgctgggtttacattactcgtcccgcgacgcgtaaacgttggtccataagggtgattgcattattacccctc
gcatgatgactgcaagcagcttcaagcgacgggtagttttcaggaaggctcgtttaaatgtacattcgcacactgactcgaaagca
gtcatgaaaaatcacaattgaagggcaatataccaaagacgataagtaggtatttttcggctagccgttgaaatcctagtaaaagccc
cgataaattaaccatctattttcacagaggcaatttagccttgtttaccttattgatcctaatactgggatccaacagttggagagtctagc
aaatgaaaatccgtccattacatgatcgtattgttgcgcgtaaaagaagaagagaccgcaactgcgggtggtatttttacc
ggcgctgcggcagaaaaaccaatcaagggtgtgtatctctgtgggtactggccgtattcttgataatggttcagtgcgaagcgctggc
ggttaacgaaggcgatgtgtcgttttggtaatactcaggtcaaaatactatcgatcgtatggtgaagaattattgattttgaatga
aagtgatctacggcggtttagaagcttaattattacactcactttttttaaactacaaaatftaaggaaagatcatggctgctaaagacg
tattatttggtagacgcacgcgcaaaaatgttggtaggtgtaaacattttagccgacgcagtaagagttaccttaggacctaa
aggctgtaacgttgttatagaaaaatcatttgggtgcaccgatcatcaccaaagatgggtgttctgttgcgcgtgaaatcgaattgaaagaca
aatcgaataacatggggcgacagatggtaagggaaggtgcttctcaagccaacgaccaagccggtagcggcacaacgacagcgactg
tactagcacaggcgattatcagcgaaggcttgaatctgttgcggctggcatgaatccaatggatcttaaacgttggtattgataaagctac
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cgatgaaacgggtggctgttaattgctgaagcgatggaaaaagtcggtaaaagaggtgtgattaccgttgaagaaggcaaaggccttg
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agcgggttaaagccctgggtttggcgatcgtcgtaaagcgatgttgcaagatcttgccatcttgacgggtggtcaggttatttctgaagag
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gttacaagaacgcgttgctaagcttgcggggcggttgcgtgattaaaggttgcgggttctgaaatggaaatgaagagagaagaaa
gaccgtgttgacgatgcacttcatgcaactcgcgcagcgggtgaagaaggtgtgttgcgggtggtggtgtgttcttgatcgcgcactct
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agtatggcgatatgattgcgatgggtatttagaccctgcaaaagtcacgcgttcattctctacaagccggcgctctatcgcaggtttgat
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cggtatgcctggcatgatgtaatcactttgtgattcattgtcctgatctgcttaccgtgtaaaagatcaggctcaaggctgtctctataaaa
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atttatgtaactagctggcctataatgttgagttcctctgggtggcatgatctcatggtacttcacttaagcctgattcactgcg
gctttaacagtaaaataataacgcaacgtagaaacataataagcgtatggcattaatgaagacggctgcatttaattcagatc